

Capillary electrophoresis of inorganic cations and low-molecular-mass amines using a copper-based electrolyte with indirect UV detection

John M. Riviello* and Michael P. Harrold

Dionex Corporation, 1228 Titan Way, Sunnyvale, CA 94088 (USA)

ABSTRACT

An electrolyte system for the separation and detection of common inorganic cations and low-molecular-mass amines using capillary electrophoresis was developed. The electrolyte system is based on copper(II) as the primary electrolyte constituent. The method permits the determination of alkali metals, alkaline-earth metals, and ammonium in less than 5 min. Utility of the method for the separation of low-molecular-mass amines has also been demonstrated. Factors affecting separations using this electrolyte system have been investigated and include pH, buffering, and the addition of selective mobility modifiers. The analytical performance of the electrolyte system is discussed in terms of detection limits, linearity of response, migration time precision and matrix effects.

INTRODUCTION

Alkali and alkaline-earth metals are monitored routinely in a variety of industries. The most common analysis methods used are spectroscopic techniques such as atomic absorption (AA) and atomic emission spectroscopy [1]. Ion chromatography has been used extensively for rapid determination of alkali and alkaline-earth metals [2–4], particularly when the metals must be separated to eliminate inter-element interferences or when ammonium ion must also be determined.

Capillary electrophoresis (CE) has recently received a great deal of attention as a tool for the separation and detection of low-molecular-mass inorganic ions. With the exception of recent research into conductivity detection for CE [5–7], most workers have used indirect methods for the detection of alkali and alkaline-earth metals. The principles of indirect UV and indirect fluorescence methods for CE are well documented

[8–13]. The separation of inorganic anions and low-molecular-mass organic acids using CE with indirect UV detection has been the subject of several recent publications [14–20]. Early work on the separation of metal ions by Hjertén [21] and Tsuda *et al.* [22] demonstrated the technique's potential for metal ion determination. Foret *et al.* [23] described the separation of lanthanide metals using indirect UV detection with a creatinine electrolyte. They used α -hydroxyisobutyric acid (HIBA) in the electrolyte to create weak complexes that enhance the small differences in electrophoretic mobility of the lanthanide metals. More recently, metal separations based on a benzylamine and HIBA electrolyte were reported by Chen and Cassidy [24]. Most work on CE of cations has used an organic amine as the primary electrolyte component and used indirect UV or indirect fluorescence detection. Several reports [25–27] describe the separation of inorganic metal ions using a proprietary compound. Inorganic-based electrolytes for the separation of metal ions have been described by Bachmann *et al.* [28] using a cerium(III)-based

* Corresponding author.

electrolyte with indirect fluorescence detection. Timerbaev *et al.* [29] separated metal ions as anionic complexes with 8-hydroxyquinoline-5-sulfonic acid and used direct UV detection. Swaile and Sepaniak [30] also used 8-hydroxyquinoline-5-sulfonic acid, but detected the fluorescent complexes of the metals.

Metal ions of a given valency in the same row of the periodic table are often difficult to separate because of very small differences in electrophoretic mobility. In many cases, the electrophoretic mobility of an ion can be changed by choosing appropriate electrolyte conditions which modify its charge, mass or charge density. Adding modifiers that complex with analyte cations to control electrophoretic mobility has been described by several workers [23,24,27]. The addition of 18-crown-6 to electrolytes in isotachopheresis has been shown to resolve ammonium and potassium ions, two species that normally co-migrate [31]. Recently, the use of 18-crown-6 in CE for the resolution of ammonium and potassium has also been described [32]. The application of CE for determining non-chromophoric amines has not been extensively investigated, although, recently, the separation of low-molecular-mass amines was reported using indirect UV detection with a quinine-based electrolyte [33].

In this paper, we report on the first CE method using an inorganic chromophore with indirect UV detection. An electrolyte system based on copper(II) has been developed for the determination of alkali and alkaline-earth metals including barium and strontium. The electrolyte system has also been used successfully for the separation and detection of low-molecular-mass amines. Optimization parameters for the method and performance characteristics are presented.

EXPERIMENTAL

Reagents and standards

All reagents were of analytical-reagent or ACS grade unless specified, and prepared in 18 M Ω deionized water. Copper(II) sulfate (pentahydrate) was obtained from MCB (Norwood, OH, USA). Formic acid was obtained from Fluka (Ronkonkoma, NY, USA) and 18-crown-6 was

from Aldrich (Milwaukee, WI, USA). Concentrated sulfuric acid was obtained from Fisher Scientific (Pittsburgh, PA, USA).

Standards for magnesium and calcium were obtained as 1000 mg/l ion standard solutions from Fluka. Standards for lithium, sodium, ammonium, potassium, strontium, and barium were prepared from chloride salts obtained from Fisher Scientific.

Amine standards were prepared from the free bases. Monomethylamine, dimethylamine, trimethylamine, monoethylamine, diethylamine and triethylamine were obtained from Fluka.

Equipment

All electropherograms were generated using a Dionex CES I system (Dionex, Sunnyvale, CA, USA). Fused-silica capillaries (Polymicro Technologies, Phoenix, AZ, USA) of 50- μ m inner diameter, 375- μ m outer diameter and various lengths were used. The detection window was located 5 centimeters from the end of the capillary. Data were collected at 10 points/s using a Dionex AI-450 data acquisition system. Unless otherwise specified, injections were performed hydrostatically by raising the sample vial 100 mm above the level of the destination vial for 30 s. To obtain positive peaks when using indirect photometric detection, the polarity of the UV-visible detector was reversed. Wavelength setting was 215 nm unless otherwise noted.

RESULTS AND DISCUSSION

Copper(II) was used as the primary component in the electrolyte because it has an electrophoretic mobility similar to that of the analytes, and also has spectral characteristics required for indirect UV detection. Cupric ion, at pH 3, has an electrophoretic mobility well matched to many inorganic cations and low-molecular-mass amines (Table I). The electrophoretic mobility of copper(II) is well matched to that of the analytes only if copper is present as a free divalent cation. Copper(II) will hydrolyze at pH greater than 5, and this hydrolysis can change its electrophoretic mobility. In order to prevent hydrolysis, the pH of the copper-based elec-

TABLE I
EQUIVALENT IONIC CONDUCTANCE (Λ) OF SE-
LECTED INORGANIC CATIONS

From ref. 34.

Metal ion	Λ ($10^{-4} \text{ m}^2 \text{ S mol}^{-1}$)
Ammonium	73.50
Potassium	73.48
Sodium	50.08
1/2 Calcium	59.47
1/2 Magnesium	53.00
1/2 Strontium	59.40
Lithium	38.66
1/2 Barium	63.66
1/2 Copper	55.00
Hydronium	349.82

trolyte is kept below 5. The electroosmotic flow rate will change as a function of electrolyte pH, and a decrease in electroosmotic flow is observed as the pH is lowered. Maintenance of a constant pH is essential for assuring reproducible migration times and minimizing the impact of injected sample pH. Formic acid, with a $\text{p}K_a$ of 3.1, was used to adjust the pH of the electrolyte to prevent hydrolysis of copper and to provide buffering capacity to the electrolyte.

The separation of ammonium and potassium using CE is difficult due to their identical electrophoretic mobilities [34]. The use of 18-crown-6 to solve this problem has been documented by other workers [31,32]. Stability constants for 18-crown-6 with selected metals are shown in Table II. Potassium ion has a stability constant with 18-crown-6 approximately six times that of ammonium. The higher stability constant for potassium results in a migration velocity that is slightly retarded relative to ammonium, and the two ions can be separated. We have examined the utility of 18-crown-6 in the copper(II)-based electrolyte system and found it to be useful for the resolution of ammonium and potassium in the copper electrolyte. Fig. 1 shows changes in migration times with the addition of 18-crown-6 to a copper(II)-based electrolyte system. While the effect on the resolution of ammonium and potassium is visible, other changes in selectivity

are also noted. Most dramatic is the large change in migration time for barium with the addition of 18-crown-6. A reversal of migration order for strontium and magnesium is also observed, although the magnitude of the change is not as great. The copper(II) electrolyte system we have investigated gave the maximum resolution of these analytes at an 18-crown-6 concentration of 3–5 mM.

The analytes of interest in this work are detected using indirect UV photometry. The use of an indirect method is required since alkali and alkaline earth metals have little or no intrinsic absorbance in the UV or visible region. An electrolyte ion used for indirect UV detection must be of the same charge as the analyte ion, possess a chromophore at a wavelength different than the analytes, and be of a low valency to enhance response. In addition to having a well-suited electrophoretic mobility for the analysis of low-molecular-mass cations, copper(II) ion fulfills the requirements for use with indirect UV detection. The wavelength at which the copper electrolyte gives the greatest signal-to-noise ratio (S/N) was determined by measuring the S/N for lithium ion at several wavelengths. The results of this experiment are shown in Fig. 2. Based on this work, all subsequent electropherograms were run at 215 nm. Using indirect UV for this application has the benefit of universal response to positively charged species. However, the method of indirect UV also has drawbacks, including a noisy, high background, relatively

TABLE II
STABILITY CONSTANTS OF METAL IONS WITH 18-
CROWN-6

From ref. 35.

Metal ion	$\log K$ at 25°C
Sodium	0.80
Potassium	2.03
Ammonium	1.23
Rubidium	1.56
Cesium	0.99
Strontium	2.72
Barium	3.87

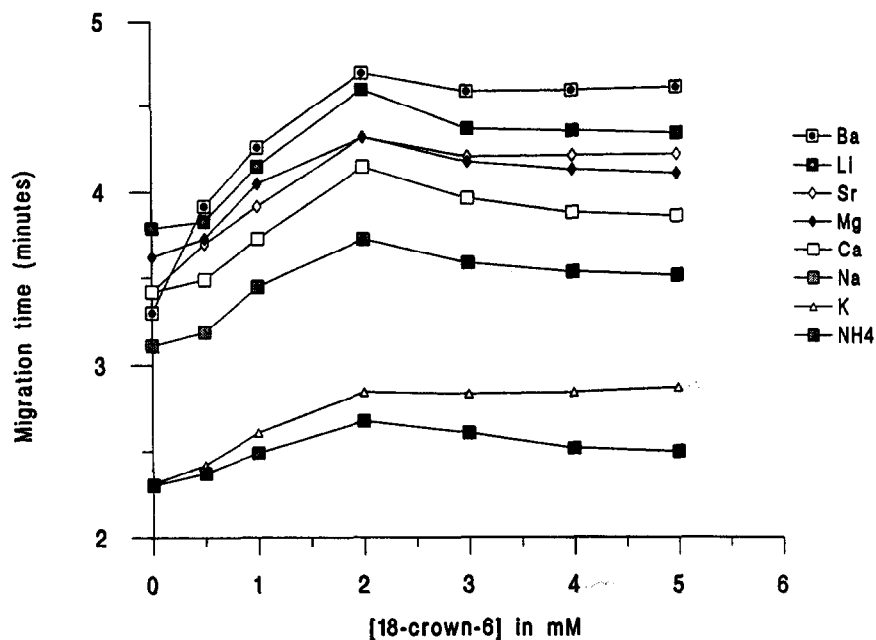


Fig. 1. Effect of 18-crown-6 on migration times of inorganic cations. Electrolyte consists of 4.0 mM cupric sulfate, 4.0 mM formic acid and varying amounts of 18-crown-6. Separation performed in 50 cm \times 50 μ m I.D. fused-silica capillary at 20 000 V, detector cathodic.

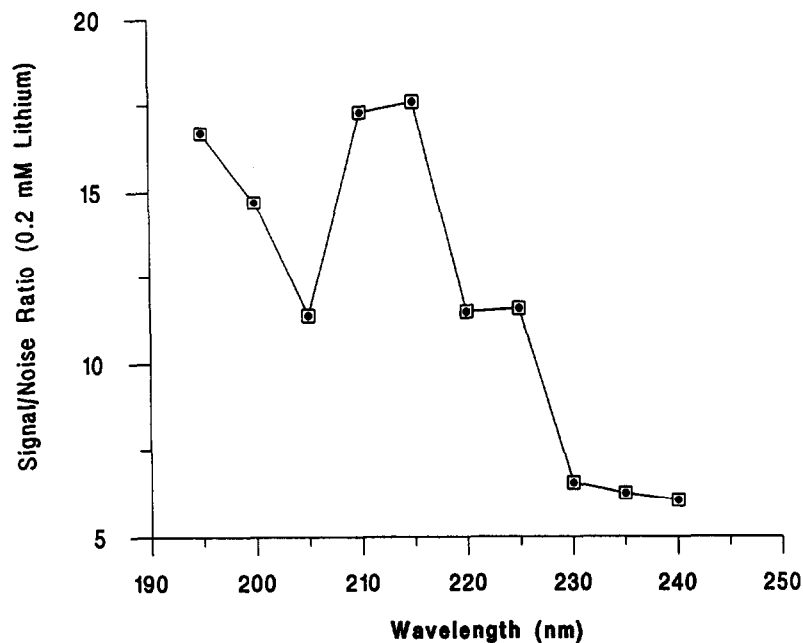


Fig. 2. Signal-to-noise ratio vs. detection wavelength for 0.2 mM lithium injected. Electrolyte consists of 4.0 mM cupric sulfate, 4.0 mM formic acid and 4.0 mM 18-crown-6 at pH 3.0.

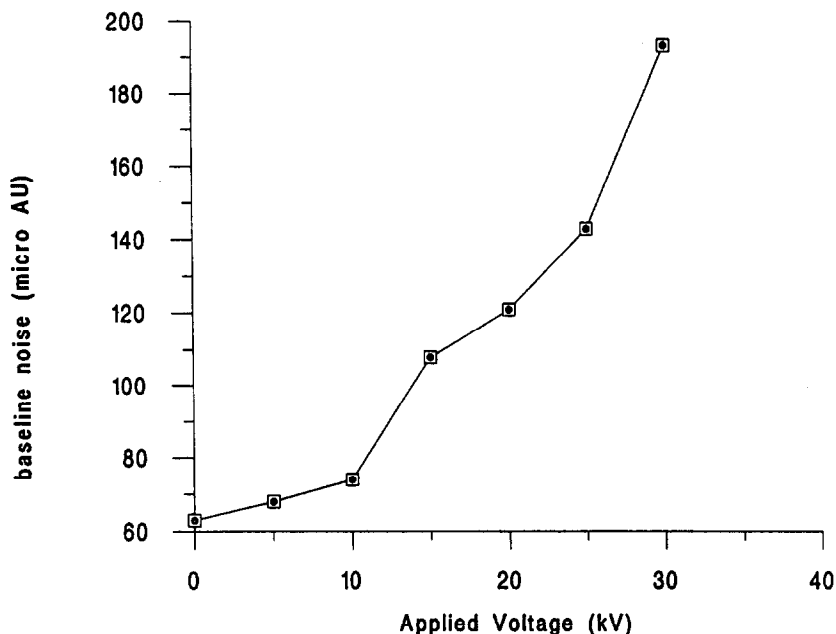


Fig. 3. Baseline noise (absorbance units $\times 10^{-6}$) as a function of applied voltage for cupric sulfate electrolyte.

high detection limits and a limited dynamic range.

Electroosmotic flow rate and analyte migration velocity are proportional to the applied voltage used for separation. The baseline noise in CE is also proportional to applied voltage. The increase in baseline noise as a function of applied voltage is particularly exacerbated when using strongly absorbing electrolytes for indirect UV detection. In optimizing the separation of cations using the copper(II) electrolyte, we have sought to minimize run time and baseline noise. Fig. 3 shows the relationship between baseline noise and applied voltage for a copper(II)-based electrolyte system. Based on these results, we have concluded that 20 000 V minimizes the time of analysis while maintaining acceptably low noise.

Performance of method

A separation using an optimized electrolyte system containing 4.0 mM copper(II) sulfate, 4.0 mM formic acid and 4.0 mM 18-crown-6 is shown in Fig. 4. Detection limits and migration time reproducibility for common inorganic cations are shown in Table III. The stated detection limits are based on a hydrostatic injection at 100

mm for 30 s. Lower detection limits may be obtained by injecting greater volumes, but often at the expense of peak efficiency and resolution.

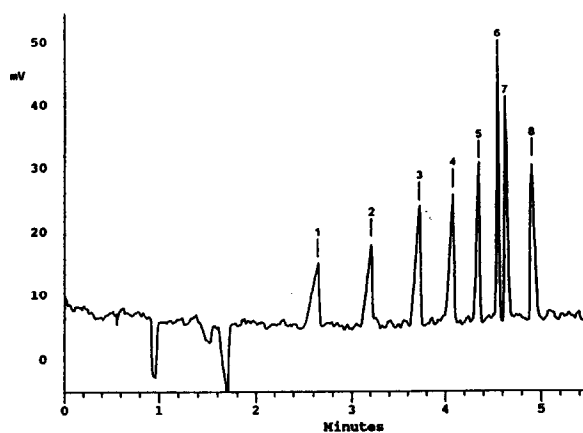


Fig. 4. Separation of alkali metals, alkaline-earth metals and ammonium. Fused silica capillary, 50 cm \times 50 μ m I.D. Electrolyte consists of 4.0 mM cupric sulfate, 4.0 mM formic acid and 4.0 mM 18-crown-6 at pH 3.0. Separation performed at 20 000 V, detector cathodic. Indirect UV detection at 215 nm, 1000 mV = 0.05 AU. Peaks: 1 = ammonium (3.6 mg/l); 2 = potassium (7.8 mg/l); 3 = sodium (4.6 mg/l); 4 = calcium (4.0 mg/l); 5 = magnesium (2.4 mg/l); 6 = strontium (15 mg/l); 7 = lithium (0.69 mg/l); 8 = barium (27 mg/l).

TABLE III
DETECTION LIMITS AND MIGRATION TIME REPRODUCIBILITY FOR CUPRIC SULFATE ELECTROLYTE

Electrolyte consists of 4.0 mM cupric sulfate, 4.0 mM formic acid and 4.0 mM 18-crown-6, pH 3.0. Separation performed in 50 cm \times 50 μ m I.D. fused-silica capillary at 20 000 V, detector cathodic. Indirect UV detection at 215 nm.

Analyte	Detection limit (mg/l) ^a	R.S.D. (%) migration time ^b
Ammonium	1.6	0.26
Potassium	1.0	0.28
Sodium	0.40	0.25
Calcium	0.33	0.26
Magnesium	0.32	0.25
Strontium	0.45	0.24
Lithium	0.060	0.25
Barium	0.80	0.28

^a Detection limit calculated as 3 \times noise using peak height.

^b R.S.D.s calculated from $n = 18$ measurements.

The linear dynamic range, measured as r^2 , for inorganic cations exceeded 0.995 from the detection limit to 50 mg/l. Loss of resolution between peaks at concentrations above 50 mg/l limited the dynamic range of each analyte. The determination of cations in drinking water using this method is shown in Fig. 5.

The separation of a series of alkyl amines is shown in Fig. 6. All the amines in this separation

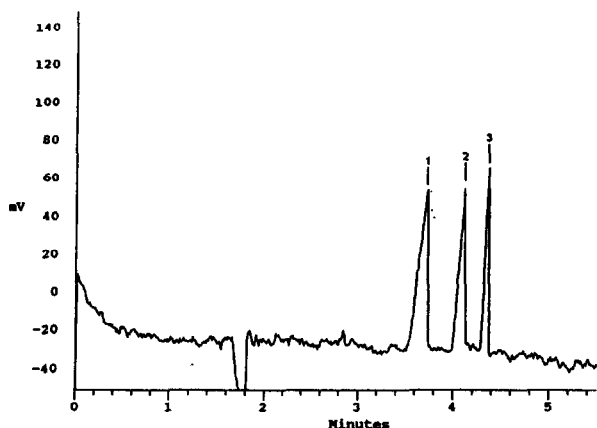


Fig. 5. Determination of cations in drinking water. Conditions as in Fig. 4. Peaks: 1 = sodium (4.9 mg/l); 2 = calcium (4.1 mg/l); 3 = magnesium (2.3 mg/l).

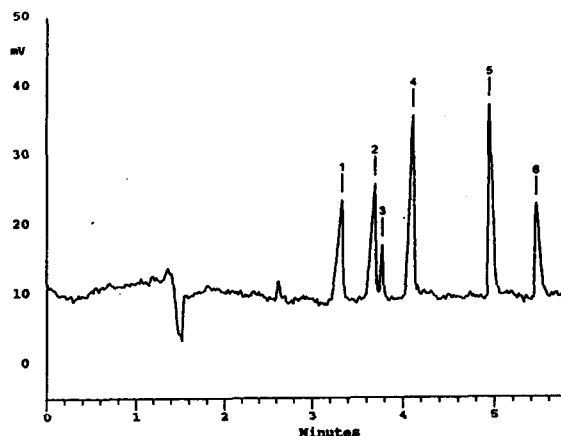


Fig. 6. Separation of amines. Conditions as in Fig. 4. Peaks (all 20 mg/l): 1 = monomethylamine; 2 = dimethylamine; 3 = trimethylamine; 4 = monoethylamine; 5 = diethylamine; 6 = triethylamine.

carry a +1 charge, and their migration from the capillary follows their electrophoretic mobilities.

The sample matrix can often cause considerable problems in separations using CE. For example, alkali and alkaline-earth metals are most stable in acidic conditions, and are often analyzed in an acidic aqueous solution. Fig. 7 shows the effect of a sulfuric acid matrix on cation migration times using a copper(II)-based electrolyte. Small amounts of acid in the sample have the effect of decreasing the migration time for all analytes. Migration times of all analytes begin to plateau as the acid concentration approaches 25 mM, but both potassium and ammonium begin to display increasing migration times above 25 mM acid. The effect is most pronounced for faster migrating species. Although changes in migration times are observed for all analytes, acid concentration as high as 25 mM in the sample will not significantly degrade the separation.

CONCLUSIONS

An electrolyte system based on copper(II) has been developed for the determination of alkali metals, alkaline-earth metals, and low-molecular-mass amines. Separation is carried out using free zone electrophoresis with indirect UV detection. Copper(II) is well suited for this application as its electrophoretic mobility is closely

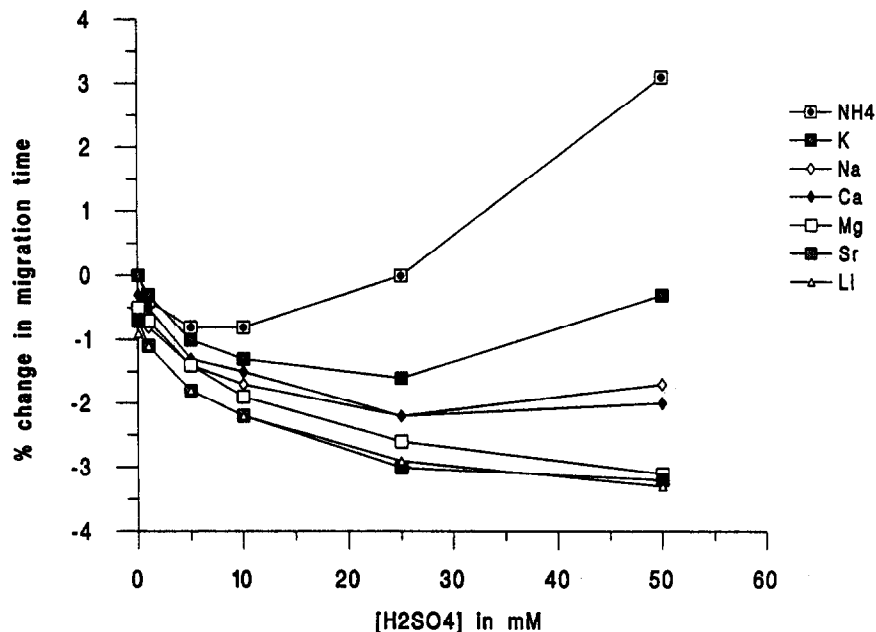


Fig. 7. Effect of sample acid concentration on migration times of inorganic cations. Electrolyte consists of 4.0 mM cupric sulfate, 4.0 mM formic acid and 4.0 mM 18-crown-6, pH 3.0. Separation performed in 50 cm \times 50 μ m I.D. fused-silica capillary at 20 000 V, detector cathodic.

matched to the analytes of interest and cupric ion has the required spectral characteristics for indirect UV detection. Resolution of otherwise co-migrating species is accomplished with the selective complexation of some species with 18-crown-6. Detection limits, linearity, and reproducibility of the method are acceptable for a variety of analytes in the 100 μ g/l to 50 mg/l range. The electrolyte system is buffered, and matrices as high as 25 mM acid may be analyzed without degrading the separation.

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